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(12) **United States Patent**
Makarov et al.(10) **Patent No.:** **US 9,410,193 B2**
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USING DNA MOLECULES GENERATED BY
RANDOM FRAGMENTATION**(71) Applicant: **RUBICON GENOMICS, INC.**, Ann Arbor, MI (US)(72) Inventors: **Vladimir L. Makarov**, Ann Arbor, MI (US); **Irina Sleptsova**, Ann Arbor, MI (US); **Emmanuel Kamberov**, Ann Arbor, MI (US); **Eric Bruening**, Chelsea, MI (US)(73) Assignee: **Rubicon Genomics, Inc.**, Ann Arbor, MI (US)

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CPC **C12Q 1/6855** (2013.01); **C12Q 1/6844** (2013.01); **C12Q 1/6846** (2013.01); **C12Q 1/6869** (2013.01)(58) **Field of Classification Search**

None

See application file for complete search history.

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The present invention is directed to methods to prepare a DNA molecule or a plurality of DNA molecules by random fragmentation. In some embodiments, the present invention regards preparing a template for DNA sequencing by random fragmentation. In specific embodiments, the random fragmentation comprises chemical fragmentation, mechanical fragmentation, or enzymatic fragmentation. In further specific embodiments, a universal sequence is attached to the 3' end of the DNA fragments, such as by ligation of an adaptor sequence or by homopolymeric tailing with terminal deoxynucleotidyltransferase. In other embodiments, a library is prepared with methods of the present invention.

17 Claims, 28 Drawing Sheets